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Reliance of coastal intertidal food webs on river input – Current and future perspectives

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ABSTRACT

Climate change models predict an important decrease in precipitation in some parts of the world, which will lead to a decrease in river plume magnitude. River plumes can have an important enhancing effect on coastal secondary production and sustain important fisheries. Studies using stable isotopes have been proven successful in characterizing the energy pathways that sustain coastal fisheries and their dependence on terrestrial matter inputs. This has been shown in subtidal coastal waters affected by major rivers, but not in the adjacent intertidal food webs. Since intertidal food webs function as nursery and refuge areas for important commercial species it is important to understand if they too are dependent on river carbon inputs. In this study, carbon and nitrogen isotopes were used to investigate the incorporation of terrestrial particulate organic matter (POM) input into the intertidal communities of the northern and southern coast adjacent to the river Tagus. In total 34 food web components were analysed. Among the top consumers, 16 fish species and two shrimp species were identified. Nine commercial fish species were analysed. Reliance on benthic affinity prey was high for most top consumers. Top consumers relied heavily on POM as a carbon source. Intertidal food webs near the Tagus river mouth are dependent on terrestrial inputs. The increase in droughts predicted by climate change models should impact these coastal food webs, since the benthic energy pathway upon which this food web relies, should be negatively impacted.

1. Introduction

Several studies have shown the importance of freshwater inputs in the enhancement of coastal secondary production (e.g. Darnaude, 2005; Dolbeth et al., 2008; Vinagre et al., 2011a,b). The terrestrial runoff carried by the river plumes of major rivers, seems to have a crucial role in sustaining important coastal fisheries in various parts of the world (e.g. Beamish et al., 1994; Grimes, 2001; Le Pape et al., 2003; Erzini, 2005; Gillson, 2011). It has been shown that nutrients carried by terrestrial run-off cause an increase in the production of coastal phytoplankton (Moutin et al., 1998; Cloern, 2001). The deposition of this phytoplankton along with the detritus carried by the terrestrial plumes leads to an increase in benthos production (Salen-Picard and Arlhac, 2002; Salen-Picard et al., 2002; Hermand et al., 2008), via the benthic detrital energy pathway, positively affecting fish that consume benthic prey (Darnaude et al., 2004; Darnaude, 2005).

Climate change is expected to have an important impact on the magnitude of river plumes, through the alteration of rain patterns in many parts of the world (IPCC, 2001, 2007, 2014, 2018). While in some areas river plumes are expected to increase, in others, such as southern Europe, they are expected to suffer an important decrease (IPCC, 2001, 2007; Miranda et al., 2002). The Portuguese coast is one of such areas, with a predicted decrease in rainfall of up to 60% (Miranda et al., 2002).

Vinagre et al (2011a), analysed the spatial variation in the influence of the river run-off along the northern subtidal area adjacent to the Tagus River, using stable isotopes and detected a river-coast gradient in the incorporation of terrestrial particulate organic matter (POM) in coastal organisms, which decreased with increasing distance from the river.

The Tagus River is one of the major rivers in Europe in terms of length and drainage. Both its northern and southern adjacent coasts support important local fisheries and function as nursery areas for several marine species (Prista et al., 2003; Dias et al., 2014, 2016). Vinagre et al (2011a) showed that, even under low flow conditions, fishing areas 30 km to the north of the river mouth were under the influence of the Tagus plume.

Given the drastic decrease in rain fall that is predicted for this region, it is important that monitoring plans are put into action in order to allow the early detection and the understanding of the potential effects

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of a reduction in the coastal enrichment by the Tagus river plume. This is particularly important since coastal secondary production sustains fishing communities, in the northern and southern coasts adjacent to the river.

Moreover, the Tagus River flow is regulated by several upstream dams, which provide flood control, drinking and farming water for more than two million people (data from Instituto Nacional de Estatística, available online at www.ine.pt). This way, it is expectable that in face of frequent and prolonged droughts, these dams will be used to contain the river drainage in a larger proportion than today.

The fact that Portugal is the third highest consumer of fish per capita in the world, that fishing is an integral part of the Portuguese social and cultural heritage, and a major means of subsistence for many coastal communities, makes this a pressing issue (Leitão et al., 2014). In this context, managers and decision-makers will need accurate information on the potential impacts of reduced river flow.

Intertidal areas may function as nursery and refuge areas for some of the most important commercial species of these area, such as various species of sea breams, mostly *Diplodus sargus* and *D. vulgaris*, sardine, *Sardina pilchardus*, and octopus, *Octopus vulgaris* (Dias et al., 2014, 2016; Moreno et al., 2014). Thus, intertidal areas are also important for the sustainability of the adjacent subtidal fisheries, however there is no knowledge on their dependence on terrestrial carbon inputs from the river Tagus. If they are shown to be as dependent as subtidal areas, then the whole coastal ecosystem can be considered as vulnerable to droughts.

The aim of the present study is to investigate the incorporation of terrestrial POM in the intertidal food webs of the northern and southern coastal zones adjacent to the Tagus River (separately), through the 1) assessment of the reliance of the consumers on benthic affinity prey and the 2) determination of the main sources of carbon used by consumers, this way providing a new and complementary insight into the reliance of the coastal food webs adjacent to the river Tagus on terrestrial inputs.

2. Material and methods

2.1. Study area

The Tagus river ends in a wide estuary, which narrows at the river mouth forming a 12 km narrow channel, with a depth of 30 m. It has a high river flow, one of the highest in Europe. Its average monthly flow ranges from 100 $m^3 s^{-1}$ to 2200 $m^3 s^{-1}$, and its average annual flow is of 300 $m^3 s^{-1}$ (Cabeçadas et al., 2000). Only 6% of the annual rainfall occurs in summer, while 42% occurs in winter. In average conditions, the Tagus river plume extends northwards along the coastline driven by currents and strong coastal winds (Vaz et al., 2009). In summer, persistent northerly winds transport the plume to the south, along the coastline (Vaz et al., 2009). The study area consists of two coastal areas adjacent to the Tagus river mouth, approximately 30 km to the north and to the south (Fig. 1).

The northern coastal area is composed mainly of rocky shores and some sandy beaches, while the southern coastal areas is a long stretch of sand, only interrupted by some man-made peers for coastal defence against winter storms. This long sandy beach ends in a rocky cape.

2.2. Sampling

Surveys were conducted in the shallow intertidal area, in late summer, September 2013, after 3 months of summer drought and low river flow. Five coastal sites were sampled at increasing distances from the river mouth, A, B and C, in the northern coast, and D and E, in the southern coast (Fig. 1). Distance between sites in the northern coast is approximately 7 km, while in the south is 19 km. A, B, C and E are located in rocky reefs, while D is located in a man-made peer.

Samples (3 replicates per site) of water (2 l per replicate), phytoplankton and zooplankton were collected at A, B, C, D and E. Water was collected in mid-water with bottles, near shore at depths of 1.20 m. Horizontal zooplankton trawls were conducted with a net of 335 µm mesh, for 5 min. Horizontal phytoplankton trawls were conducted with a net of 55 µm mesh, for 5 min. These trawls were manually performed in midwater, at a depth of approximately 1.20 m. Macroalgae and intertidal animals were collected by hand or with hand nets. Fish occurring in tide pools or shallow channels were caught with hand nets or with a fishhook. Three replicates of macroalgae and 8 replicates of animals were collected at each site.

Three water samples for POM analysis (2 l per replicate) were also collected at two sites at the river mouth, both in the north and in the south bank (Fig. 1), they will allow the isotopic determination of the water POM exported from the estuary; in the Tagus river, 30 km upstream, where river water enters the estuary (site R, Fig. 1), and in the coast, 80 km North of the estuary, allowing the isotopic determination of the marine signature (site M, Fig. 1).

2.3. Isotopic analysis

Stable isotopic analysis is effective for the characterization of spatial gradients of incorporation of material of terrestrial origin in coastal food webs (Darnaude et al., 2004; Kostecki et al., 2010; Vinagre et al., 2011a, 2012). Terrestrial primary producers usually have lower $\delta^{13}C$ than marine producers (Haines and Montague, 1979; Riera and Richard, 1996; Bouillon et al., 2000), while the increment of $\delta^{13}C$ from prey to predator is only 0–1‰ (De Niro and Epstein, 1978; Post, 2002), deeming this isotope very useful for the identification of the origin of the primary source of the organic carbon in a transitional ecosystem (Simenstad and Wissmar, 1985; Paterson and Whitfield, 1997). The nitrogen isotope informs on the trophic level, because $\delta^{15}N$ suffers an increment of 2.5–4.5‰ from prey to predator (Peterson and Fry, 1987; Post, 2002). It also informs about contamination of anthropogenic origin, since humans occupy top levels in the food chains (Hansson et al., 1997; Risk and Erdman, 2000; Gaston and Suthers, 2004).

Samples of water for POM analysis were filtered onto pre-combusted filters. Samples of phytoplankton and zooplankton were frozen and dried at 60 °C. For cnidarian species the whole body was used after their stomachs were emptied. For molluscs, the foot was dissected. Tissues of all species were left to dry at 60 °C and ground to powder.

When there was a suspicion that the samples could have carbonate contamination, such as in POM, phytoplankton, zooplankton and invertebrate species, a subsample was collected and acidified with 10% HCl while being observed under a stereoscope, while the other subsample was kept unacidified. When bubbling occurred, the whole sample was acidified, rinsed off with distilled water, and left to dry again at 60 °C, as described by Pinnegar and Polunin (1999). High carbonate content and low organic matter content in the sediment made extraction of the sediment organic matter impossible and thus the analysis was not carried out.

The ratios of $^{13}C/^{12}C$ and $^{15}N/^{14}N$ were determined by continuous flow isotope mass spectrometry (CF-IRMS) (Preston and Owens, 1983), on a Isoprime (GV, UK) stable isotope ratio mass spectrometer, coupled to an EuroEA (EuroVector, Italy) elemental analyser for online sample preparation by Dumas-combustion. IAEA-N1 and IAEA-600 were used as standards for the nitrogen isotope ratio, and IAEA-CH6 and IAEA-CH7 or IAEA-600 for carbon isotope ratio; $\delta^{15}N$ results were referred to Air and $\delta^{13}C$ to PeeDee Belemnite (PDB). Precision of the isotope ratio analysis, calculated using values from 6 to 9 replicates of laboratory standard material interspersed among samples in every batch analysis, was ≤ 0.2‰.

Isotope ratios were expressed as differences in parts per thousand (‰) in relation to a standard reference material:

$$\delta X = [(R_{sample}/R_{standard}) - 1] \times 10^3,$$

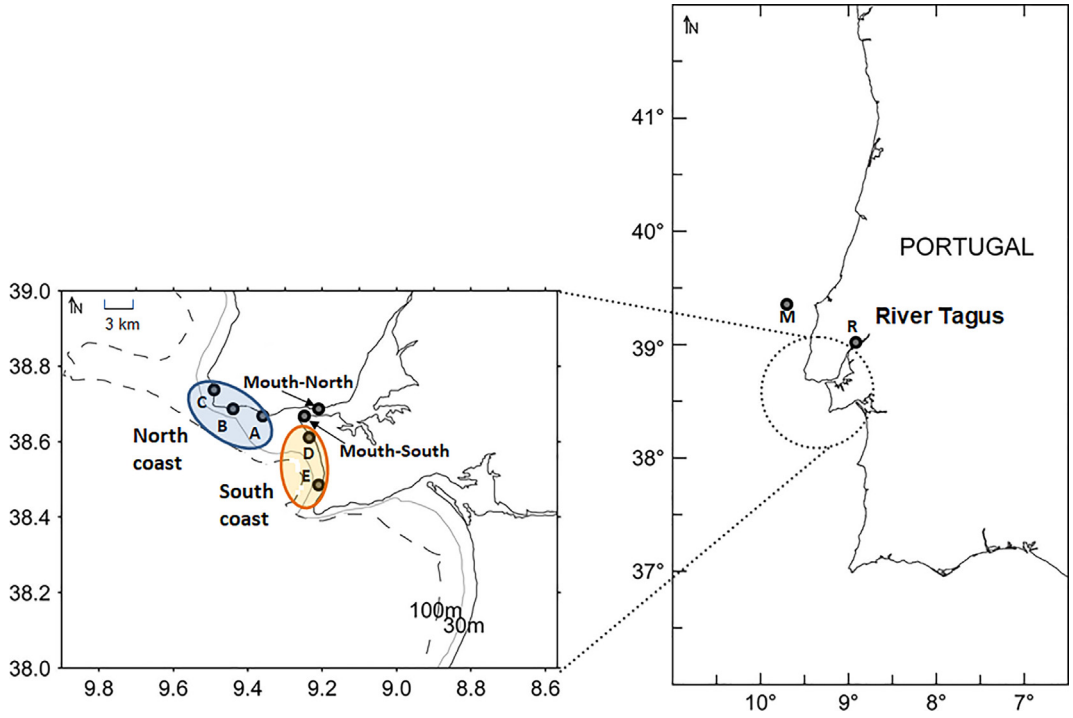

Fig. 1. Study area and sampling sites along the Portuguese coast (A, B, C, D, E, Mouth-North, Mouth-South, R and M).

where X is ^{13}C or ^{15}N, R is the ratio of $^{13}C/^{12}C$ or $^{15}N/^{14}N$ and δ is the measure of heavy to light isotopes in the sample.

2.4. Data analysis

Differences in $\delta^{13}C$ and $\delta^{15}N$ among the local water POM reservoirs (sites A, B, C, D and E) and main POM sources (sites R, Mouth-north, Mouth-south and M) were analysed separately using an ANOVA, followed by Tukey post-hoc tests. The ANOVA assumptions were previously investigated. A significance level of 0.05 was considered.

Trophic level (TL) was estimated according to the following equation:

$$TL = (\delta^{15}N_{consumer} - \delta^{15}N_{baseline})/3.4 + 1$$

where 3.4‰ is the ^{15}N trophic fractionation factor, following Minagawa and Wada (1984), Post (2002) and Akin and Winemiller (2008). $\delta^{15}N$ mean baseline was a mean of the isotopic values of macroalgae, benthic isotopic signature (POM) and pelagic isotopic signature (*Mytilus galloprovincialis*, after substraction of 3.4 to account for trophic level fractionation). Three end-members were used for this calculation because there is no prior knowledge on which one is dominant at the study area.

The reason for using *M. galloprovincialis* (minus trophic fractionation) and not phytoplankton directly was because this is a coastline affected by upwelling that is temporally and spatially very variable. This way a punctual phytoplankton sample does not incorporate the isotopic variation that occurs in the area affecting the pelagic end-member. A phytoplankton filter-feeder, such as *M. galloprovincialis* (Riisgård et al., 2011) will integrate this variability in its tissues over time, thus being a more reliable end-member (after subtraction of the trophic enrichment value).

Reliance on benthic affinity prey (RBAP) of secondary consumer species was estimated, following the equation of Le Loc'h et al. (2008):

$$RBAP = (X + (\delta^{13}C_{consumerII} - \delta^{13}C_{consumerI\ Pelagic\ affinity\ baseline})) / (\delta^{13}C_{consumerI\ Benthic\ affinity\ baseline} - \delta^{13}C_{consumerI\ Pelagic\ affinity\ baseline}) \times 100,$$

where $\delta^{13}C_{consumerI\ Benthic\ affinity\ baseline}$ and $\delta^{13}C_{consumerI\ Pelagic\ affinity\ baseline}$ are the mean of the benthic and pelagic primary consumer used as baseline. Here, *Mytilus galloprovincialis* was considered as the pelagic primary consumer baseline (because it is a well-known filter-feeder) and *Phorcus lineatus*, *Gibulla umbilicalis* and *Patella depressa* as the benthic primary consumer baseline (mean values were used). To test if the pelagic consumer and the benthic primary consumers' $\delta^{13}C$ was significantly different, a *t*-test was performed. A significance level of 0.05 was considered. The gastropods collected were selected as a reference because they are deposit-feeders/micrograzers (Hawkins et al., 1989). Primary consumers are commonly used as isotopic baseline (e.g. Post, 2002; Le Loc'h et al., 2008; Vinagre et al., 2015). $\delta^{13}C_{consumerII}$ is the $\delta^{13}C$ of the secondary consumer considered. RBAP is calculated for consumers with TL higher than 2. *X* represents the trophic enrichment along the food web, as in the equation:

$$X = -(TL_{consumerII} - TL_{consumer\ I\ Benthic\ affinity\ baseline}),$$

where $TL_{consumer\ I\ Benthic\ affinity\ baseline}$ and $TL_{consumerII}$ are, respectively, the trophic level of the benthic primary consumer baseline and secondary consumer.

In order to estimate the relative importance of potential primary organic matter sources to the diet of the consumers sampled, IsoSource mixing models (Phillips and Gregg, 2003) were applied to each top consumer in the north and in the south coasts' food webs (separately). The IsoSource model examines all possible combinations of each source contribution (0–100%) in small increments of 1%. Combinations that sum to the observed mixture isotopic signatures within a tolerance interval were considered to be feasible solutions. Tolerance was initially set at 0.1‰ and incrementally increased by 0.1‰ up to a maximum of 0.5‰, as suggested by Benstead et al. (2006). The criteria for choosing this model was its ability to provide results without high data requirements (Layman et al., 2012). This makes this model particularly useful when applying stable isotopes to a previously unstudied area. In case-studies where the variability of the isotopic signatures is already known more advanced models can be used, such as Bayesian mixing models.

The distribution of feasible solutions was given as a range from the

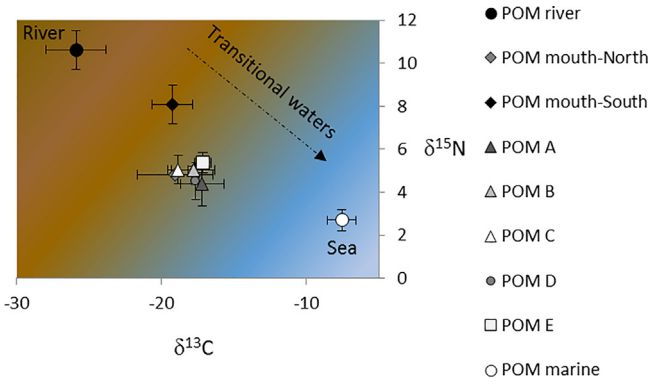

Fig. 2. Isotopic signatures of water particulate organic matter (POM) at sites A, B, C, D, E, Mouth-North, Mouth-South, at the river Tagus site (R) and at the marine site (M).

1st percentile to the 99th percentile. To account for nitrogen fractionation, 3‰ was subtracted per trophic level from the nitrogen isotope signature of the consumers (De Niro and Epstein, 1978; Minagawa and Wada, 1984). Fractionation of $\delta^{13}C$ is negligible (Peterson and Fry, 1987), so no adjustment was necessary for this isotope. Values used as sources can be seen in the Supplementary Material (MS1). The value used for algae is a mean value for all macroalgae species sampled in the area.

3. Results

As expected the $\delta^{13}C$ values of POM increased from the river (site R) to the marine site (M), assuming intermediate values in the coastal transitional waters, while $\delta^{15}N$ decreased along the same river-sea gradient (Figs. 2 and 3). Significant differences were detected for the POM of the different sites and sources, for both $\delta^{13}C$ ($F = 8.93$, $p < 0.05$) and $\delta^{15}N$ ($F = 6.39$, $p < 0.05$).

In total 34 food web components were isotopically characterized, 28 components occurred in the north coast and 19 in the south coast (eleven of the components were the same in both coastal areas) (Fig. 4). Among the consumer species, 23 were analysed in the north and 15 in the south (Supplementary material - Table 1 and 2). The maximum TL registered in the north coast was 3.8, for the goby *P. minutus*, and 3.9 in the south coast, for the seabream *D. vulgaris* (Table 1). Most of the top consumers (TL > 3.0) were fish, with the exception of two Palaemon shrimp, *P. elegans* and *P. serratus*, and the anemones *A. fragacea* and *A. equina* and *A. sulcata* (Table 1).

The most represented taxa were fish, with 20 species, ranging in TL from 2.3 to 3.9 and thus encompassing primary to secondary consumers (Table 1). Only two crustaceans were analysed, the shrimp *P. elegans* and *P. serratus* (TL = 3.0, Table 1). The cnidarians analysed were common intertidal anemones (TL = 2.2–2.7, Table 1). Molluscs were represented by one bivalve, *M. galloprovincialis*, and three gastropods, *P. lineatus*, *G. umbilicalis* and *P. depressa*.

Reliance on benthic affinity prey (RBAP) was high in both coasts, with maximum values of 94% in the north (0–94%) and 73% in the south. RBAP was above 50% for six out of 18 top species in the north, and four out of 11 top species in the south coast (Table 1).

The mixing models revealed that all fish with TL > 3.0 relied heavily on POM as a carbon source (> 50%) (Table 2). In fact, only two of the species analysed did not rely mostly on POM, these were the two shrimp species, *P. elegans* and *P. serratus* (Table 2). Shrimp depends on phytoplankton and POM to a considerable extent (37–48%). The role of macroalgae as potential sources of carbon for the top consumers of both food webs was very small (Table 2).

4. Discussion

4.1. Food web structure

This study characterised 34 components of an intertidal food web, a considerable higher number than previously analysed. Vinagre et al. (2015) reported only 19 food web components in an isotopic study covering a much wider area of the Portuguese coast (encompassing a coastal stretch approximately 30 km north of this study area, up to 100 km further north). The intertidal coast investigated by Vinagre

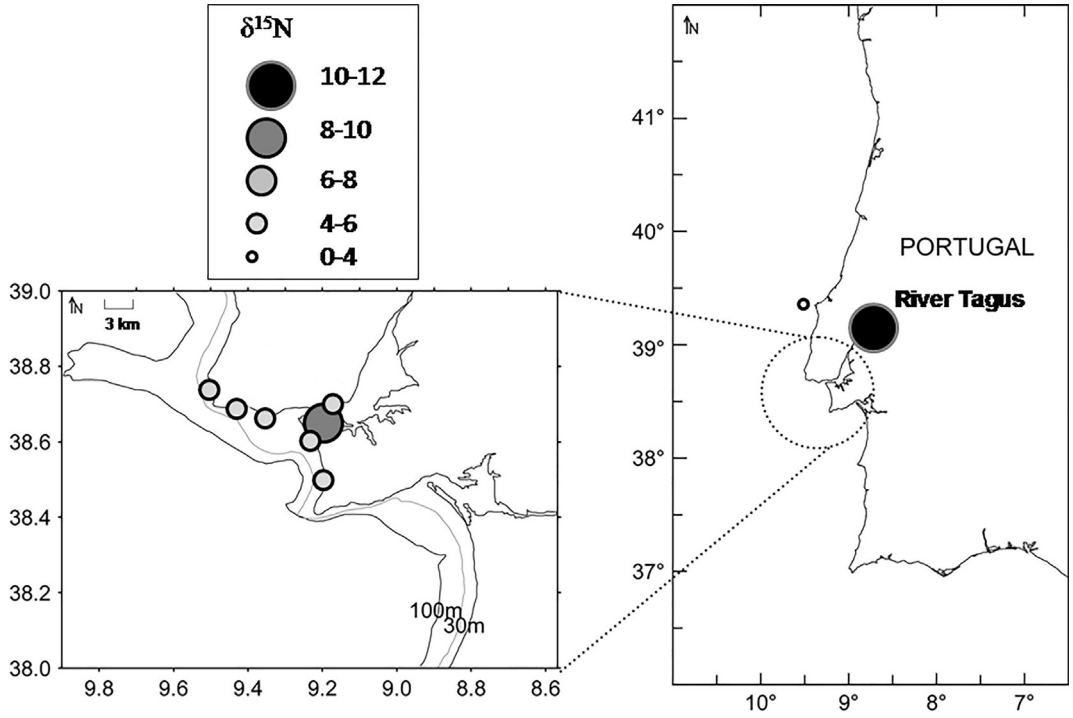

Fig. 3. Values of $\delta^{15}N$ registered for the particulate organic matter of water, at each site.

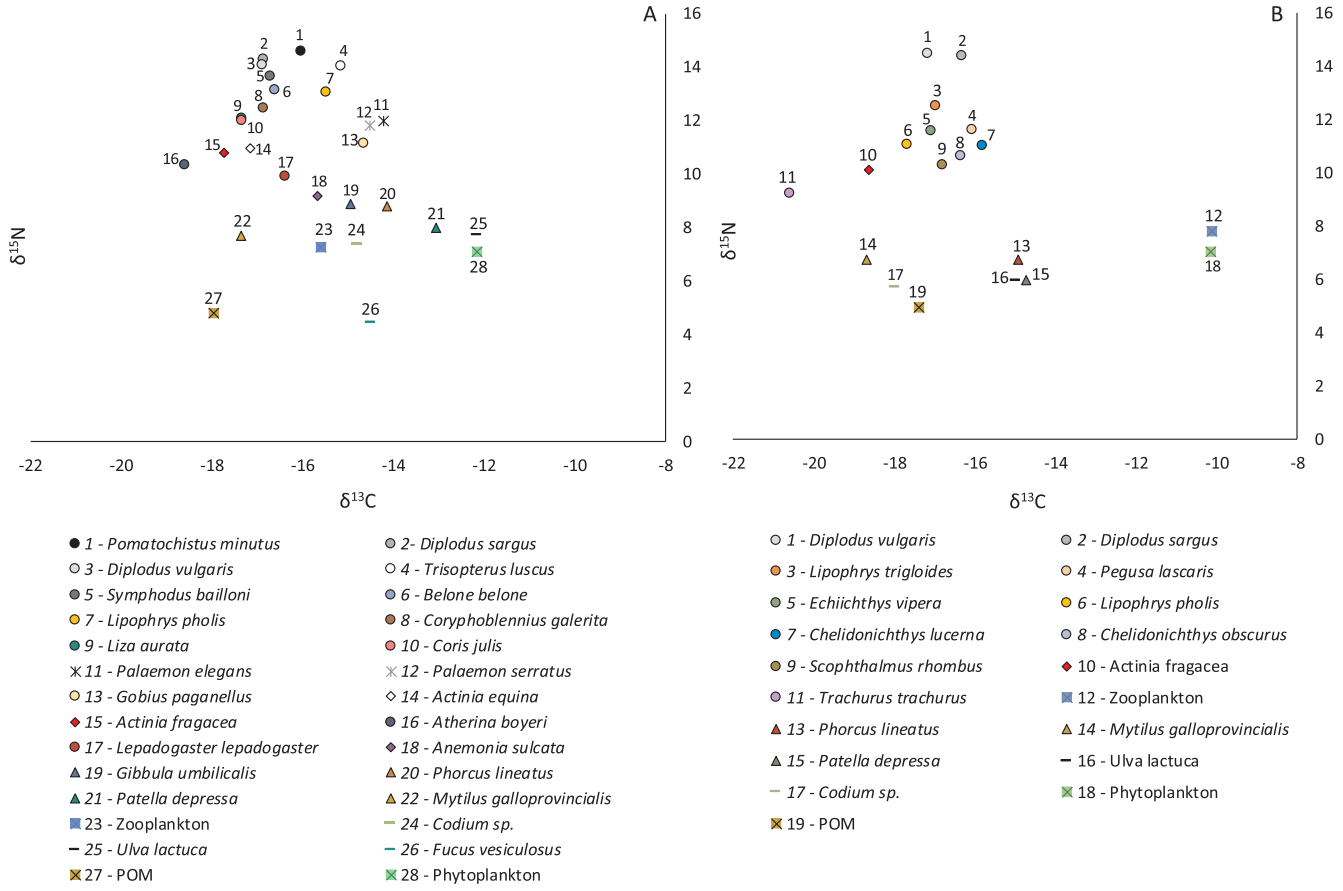

Fig. 4. Mean isotopic values of each food web components analysed at the north (a) and the south coast (b).

Table 1
Trophic level (TL) of the consumers (TL > 2.0) and reliance on benthic afinity prey index (RBAP) in the north and south coasts. Organisms used as baseline are not included. Values of RBAP > 50 are presented in bold.

Species	North		South	
	TL	RBAP	TL	RBAP
Pomatochistus minutus	3.8	39		
Diplodus sargus	3.7	13	3.8	**59**
Diplodus vulgaris	3.6	13	3.9	37
Trisopterus luscus	3.6	**66**		
Symphodus bailloni	3.5	18		
Belone belone	3.3	22		
Lipophrys trigloides			3.3	43
Lipophrys pholis	3.3	**56**	2.9	25
Pegusa lascaris			3.0	**67**
Echiichthys vipera			3.0	40
Coryphoblennius galerita	3.1	14		
Chelidonichthys lucerna			2.9	**73**
Chelidonichthys obscurus			2.7	**60**
Liza aurata	3.0	0		
Coris julis	3.0	0		
Scophthalmus rhombus			2.6	48
Palaemon elegans	3.0	**94**		
Palaemon serratus	3.0	**85**		
Actinia fragacea	2.6	0	2.6	0
Gobius paganellus	2.7	**81**		
Actinia equina	2.7	6		
Atherina boyeri	2.5	0		
Trachurus trachurus			2.3	0
Lepadogaster lepadogaster	2.4	0		
Anemonia sulcata	2.2	**51**		

et al. (2015), albeit similar to the one studied here, was not under the influence of any major river.

Similarly, to what had been reported for other intertidal and subtidal ecosystems in Portugal (Vinagre et al., 2012, 2015) and in other parts of the world (e.g. Darnaude et al., 2004; Le Loc'h et al., 2008), this food web was generally composed by fish as top consumers, followed by shrimp at an intermediate level, anemones as lower secondary consumers and molluscs as primary consumers. This general structure was also found in similar isotopic studies conducted in subtidal estuarine ecosystems (e.g. Vinagre et al., 2008, 2011a,b; Kostecki et al., 2010; Pasquaud et al., 2010).

Food web length was 3.9, higher than found by Vinagre et al. (2015) for a nearby intertidal coastal stretch with no major river inputs, which may indicate that the extra input in terrestrial carbon received by intertidal ecosystems adjacent to major rivers influences the length of their food webs. Subtidal waters under the influence of the Tagus river have been shown to have a shorter food web, reaching a maximum TL of only 2.4 (Vinagre et al., 2012). Other authors, using similar methods, reported longer food webs for subtidal coastal ecosystems in the Bay of Biscay, with a maximum TL of 4.1 (Le Loc'h et al., 2008), in Artic waters, with a maximum TL of 4.6 (McMeans et al., 2013), and for an artificial reef in the Mediterranean, with a maximum TL of 4.9 (Cresson et al., 2014).

Values for $\delta^{13}C$ in phytoplankton were considerably higher than previously reported for subtidal ecosystems in the same area (Vinagre et al., 2011a,b), for intertidal ecosystems with no river influence just north of the study area (Vinagre et al., 2015) and in other coastal systems (e.g. Liénart et al., 2018). Such values were also not in line with the isotopic values of various consumers of the food web of the study area. We believe that this results from the highly variable hydrographic conditions of the area, which is affected by very variable upwelling,

Table 2
Feasible percent contributions (1st and 99th percentile) of potential carbon sources to the top intertidal consumers (TL ≥ 3.0), as determined by the IsoSource model for the North and South coast (- stands for species absence at the site, POM – stands for particulate organic matter, PhytoP. – stands for phytoplankton, ZooP.- stands for Zooplankton, Algae – stands for *macroalgae*, major contributors (that contribute more than 50%) are represented in bold).

Consumer species	North	South
P. minutus	**POM 71** PhytoP. 29 Algae. 0	-
D. sargus	**POM 83–86** PhytoP. 14–15 Algae 0–3	**POM 71–96** PhytoP. 0–4 Algae 0–29
D. vulgaris	**POM 88** PhytoP. 12 Algae 0	**POM 88** PhytoP. 12 Algae 0
T. luscus	**POM 55–59** PhytoP. 41–43 Algae 0–4	-
L. trigloides	-	POM 0–15 PhytoP. 0–2 **Algae 83–100**
S. bailoni	**POM 65–77** PhytoP. 19–23 Algae 0–16	-
B. belone	**POM 76–77** PhytoP. 22–23 Algae 0–2	-
L. pholis	**POM 63–64** PhytoP. 0–36 Algae 0–1	**POM 98–100** PhytoP. 0 Algae 0–2
P. lascaris	-	**POM 61–82** PhytoP. 14–19 Algae 0–25
E. vipera	-	**POM 53–74** PhytoP. 0–3 Algae 23–47
C. galerita	**POM 73–81** PhytoP. 18–22 Algae 0–9	-
C. lucerna	-	**POM 82–84** PhytoP. 15–16 Algae 0–3
L. aurata	**POM 75–87** PhytoP. 9–13 Algae 0–16	-
C. obscurus	-	**POM 85–95** PhytoP. 4–9 Algae 0–11
C. julis	**POM 61–74** PhytoP. 7–12 Algae 14–32	-
S. rhombus	-	**POM 100** PhytoP. 0 Algae 0
P. elegans	POM 37–44 **PhytoP. 56–61** Algae 0–7	-
P. serratus	POM 47–48 **PhytoP. 51–52** Algae 0–2	-

both temporally and spatially (Huthnance et al., 2002) and also by river runoff that is highly influenced by tides and wind conditions (Vaz et al., 2009; Valente and Silva, 2009). Such highly variable conditions should be quickly reflected by isotopic values in small-short-lived organisms, such as the one that compose plankton, while larger organisms should integrate the variation over larger time-scales in their tissues with much longer isotopic turn-over periods.

4.2. Reliance on benthic affinity prey

The reliance on benthic affinity prey is an index that illustrates the potential of each top consumer to use the benthic energy pathway as its major food source. The fact that a top consumer has a high reliance on prey of benthic affinity does not directly indicate that it incorporates organic matter of terrestrial origin, however such prey commonly relies on detritus, which in turn receives extra inputs under the influence of a major river.

Reliance on benthic affinity prey (RBAP) was high in both coasts, it was above 20% for 8 out of 18 top species in the north, and 9 out of 11 top species in the south coast. RBAP was higher than 50% for some important commercial fish species, the seabream *D. sargus*, the pouting, *T. luscus*, the sand sole, *P. lascaris*, the gurnards, *Chelidonichthys lucerna* and *C. obscurus*, the non-commercial but important tide pool fishes, the blenny *Lipophrys pholis* and the goby *Gobius paganellus*, the two shrimps, *P. elegans* and *P. serratus*, and the anemone *A. sulcata*. This indicates that this food web relies on the benthic energy pathway, which could mean that alterations in the amount of detritus due to a decrease in river plume input should cause important effects in the energy flowing from the bottom to the top of the food web.

Part of the benthic energy pathway is maintained by macroalgal production, which could be increased by global change, due to higher temperatures, CO_2 and nutrient availability, somewhat counterbalancing the loss of riverine POM. Knowledge on how this may compensate the decrease in river input will require in-depth ecological modelling of the complex processes involved in benthic–pelagic coupling (Griffiths et al., 2014).

The seabream *D. sargus*, the blenny *Lipophrys pholis* and the two shrimps, *P. elegans* and *P. serratus*, had previously been reported as species with a high RBAP (> 50%), in the intertidal areas with no river influence (Vinagre et al., 2015). This shows that even when the additional input of river carbon is not present, species do not change their main prey and still rely on prey with a benthic affinity.

4.3. Role of river POM as energy source

Water POM of each coastal site presented isotopic values ranging between those of the Tagus river and marine waters, indicating that these are the two main sources of carbon and nitrogen to the coastal areas studied, as previously observed by Vinagre et al. (2011a).

The decreasing gradient in $\delta^{15}N$ values observed in this study, from the river to the sea (Fig. 3), were also previously reported (Vinagre et al., 2011a). This gradient is typical of coastal waters under the input of polluted river waters (Cromey et al., 1998; Alonso-Rodriguez et al., 2000; Riera et al., 2000). Humans are high in the food chain, consequently their sewage is enriched in $\delta^{15}N$, making this isotope a marker for anthropogenic contamination (Hansson et al., 1997; Risk and Erdman, 2000; Gaston and Suthers, 2004). In fact, $\delta^{15}N$ has been successfully used to map and trace the input of anthropogenic sewage in other coastal areas (Costanzo et al., 2001; Waldron et al., 2001; Parnell, 2003; Gaston et al., 2004).

Tagus river $\delta^{15}N$ values observed in 2013 (present study) were within the range of values found in 2009 (Vinagre et al., 2011a) and those detected in other areas receiving human sewage effluents in Australia (e.g. Costanzo et al., 2001; Gaston et al., 2004; Hadwen and Arthington, 2007) and Scotland (Waldron et al., 2001). The current values for the Tagus river were considerably higher than that found in 2001 (Vinagre et al., 2008), which was 4‰. This may be an effect of the effort in sewage treatment that took place in the past decade. Sewage treatment encompasses microbial degradation of organic matter, which adds a fractionation step to the $\delta^{15}N$ values, possibly causing the higher values detected since 2009.

The application of mixing models to estimate the feasible contributions of the potential carbon sources to the top intertidal consumers revealed that they depend heavily on coastal POM. Since coastal POM in these areas is composed of a mix of local and riverine POM, it may be concluded that the top consumers of these food webs are dependent on carbon of river origin and should thus be negatively affected

if that carbon input decreases due to the more intense, frequent and prolonged droughts predicted by climate change models. Results from the reliance on benthic affinity prey (RBAP) complement these results, showing that the benthic energy pathway, that includes local and riverine detritus, is high for many consumers.

Mixing models also showed that, in the food webs analysed here, all of the fish that were top consumers (16 species) (TL ≥ 3.0) were dependent on POM as their major carbon source (> 60%). Among these 16 fish species, there are 9 commercial species, encompassing seabreams (*D. sargus* and *D. vulgaris*), sand sole (*P. lascaris*), pouting (*T. luscus*), garfish (*B. belone*), gunards (*C. lucerna* and *C. obscurus*), wrasse (*Coris julis*) and brill (*Scophthalmus rhombus*). The only two top consumers that are not fish, were the two shrimp species, which depended mostly on phytoplankton, but still depended on POM to a considerable degree, 37–48%.

5. Conclusions

This study showed that the intertidal food webs of the coasts adjacent to the river Tagus, present a food web with a maximum length of 3.9, higher than observed in similar areas not influenced by major rivers. The food web structure was dominated by coastal fish, assuming top consumer roles, followed by crustaceans, anemones and molluscs as lower level consumers, similarly to other intertidal and subtidal food webs. The top consumers of these food webs rely heavily on prey of benthic affinity and have POM as their major carbon source, placing them as highly vulnerable to alterations in the benthic energy pathway feeding the coastal ecosystem.

The decrease in detritus caused by droughts would create a deficit in the usual amount of organic matter that enters the benthic energy pathway, negatively affecting organisms that profit from such carbon input: detritivores, suspension-feeders and their predators, potentially cascading to the top levels of the food web, as shown by the high reliance on benthic affinity prey showed by the top predators analysed here. This way, climate change may lead to important alterations in these intertidal food webs. It can also be concluded that the intertidal food webs of the coastal areas adjacent to the river Tagus are just as dependent on river input as previously shown for their subtidal areas, and that this way the coastal ecosystem as a whole should be affected by droughts.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ecolind.2019.01.064.

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